1/3

Figure 1: Genomic constitution of certain Brassica species (U, 1935).

Amphidiploids listed in bold text

Brassica rapa Diploid Genome - AA

Brassica napus Brassica juncea Amphidiploid Amphidiploid Genome - AACC Genome - AABB

Brassica oleraceae Brassica nigra Diploid Diploid Genome - CC Genome - BB

> Brassica carinata Amphidiploid Genome - BBCC

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Reply to Office Action of August 31, 2006
New Sheet

2/3

Figure 2: Breeding procedure used to develop herbicide tolerant

Brassica juncea

46A72

Males

Males

Females Male

Bulk population from 16 Brassica juncea breeding lines low glucosinolate (9-18 umoles)

low glucosinolate (9-18 umole low erucic acid (<1%)

Female

Crossed to produce the F1
Female

F1 from previous cross Bulk pollen from 16 breeding lines –

13 F1 lines x 15 plants per line F5 to F8 generation Selected with Pursuit® 50ml/ha a.i. low glucosinolate (<8 um) Chose resistant plants for crossing low erucic acid (< 0.5%)

Crossed to produce BC1

Female Males

BC1 populations from previous cross Bulk pollen from 16 breeding lines – 6 BC1 populations x 36 plants per line F5 to F8 generation

Selected with Pursuit® - 50 ml/ha a.i. low glucosinolate (<8 umoles)
Chose resistant plants for crossing low erucic acid (<0.5%)

Crossed to produce BC2

BC2 seed from previous cross Bulk pollen from 3 breeding lines –

BC2 seed from previous cross
4 BC populations
Bulk pollen fro
F6 generation

Selected with Pursuit® – 50 ml /ha a.i. low glucosinolate (6 to 12 umoles)

Chose resistant plants for crossing low erucic acid (<0.5%)

Crossed to produce BC3

Stable juncea phenotype combined with Pursuit® tolerance Lines coded: 98SJ-23841, 98SJ-23844, 98SJ-23845

3/3

Figure 3: Greenhouse and field evaluation of Herbicide tolerant
Brassica juncea populations

Greenhouse evaluation 1 - verify tolerance and juncea phenotype

98SJ-23841, 98SJ-23844, 98SJ-23845 and unstable BC3 sister lines planted for herbicide tolerance evaluation

Pursuit® applied at 50 ml/ha a.i.; juncea phenotype stable

Survivors self pollinated and harvested

Greenhouse evaluation 2 - verify tolerance and juncea phenotype

Survivors from previous project planted for herbicide tolerance evaluation

Pursuit® applied at 50 ml/ha a.i.; juncea phenotype and tolerance stable in 98SJ-23841, 98SJ-23844 and 98SJ-23845

Survivors self pollinated and harvested

Field evaluation 1 - verify tolerance and juncea phenotype under field conditions

Pioneer Hi-Bred International Puerto Vallarta Mexico Research Station

Self-pollinated selections from all other previous projects were planted at a single location

Odyssey® was applied at 30g/ha a.i.

Juncea phenotype stable – tolerance present in 98SJ-23841, 98SJ-23844 and 98SJ-23845 progenies

Other material derived from other generations and breeding lines exhibited a range of tolerance ranging from fully resistant, intermediate resistant and susceptible. Plant phenotypes ranged from full Brassica napus to Brassica juncea phenotypes and lines and populations that exhibited traits that were intermediate between Brassica napus and Brassica juncea. In these other materials, full resistance to the herbicide was not associated with the juncea phenotype, and vice-versa.